

SEQUENCE LISTING

<110> Short, Jay M.
 Kretz, Keith A.
 Gray, Kevin A.
 Barton, Nelson Robert
 Garrett, James B.
 O' Donoghue, Eileen
 Mathur, Eric J.

<120> RECOMBINANT PHYTASES AND METHODS OF MAKING AND USING THEM

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<141> 2003-06-20

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<151> 2001-05-24

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 15 20 25 30

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 Glu Ser Val Val Ile Val Ser Arg His Gly Val Arg Ala Pro Thr Lys
 35 40 45

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 Ala Thr Gln Leu Met Gln Asp Val Thr Pro Asp Ala Trp Pro Thr Trp
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 Pro Val Lys Leu Gly Trp Leu Thr Pro Arg Gly Gly Glu Leu Ile Ala
 65 70 75

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 Page 6

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Leu	Thr	Glu	Ile	Phe	Leu	Leu	Gln	Gln	Ala	Gln	Gly	Met	Pro	Glu	Pro	
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ggg	tgg	gga	agg	atc	acc	gat	tca	cac	cag	tgg	aac	acc	ttg	cta	agt	997
Gly	Trp	Gly	Arg	Ile	Thr	Asp	Ser	His	Gln	Trp	Asn	Thr	Leu	Leu	Ser	
255					260				265						270	
ttg	cat	aac	gcg	caa	ttt	tat	ttg	cta	caa	cg	acg	cca	gag	gtt	gcc	1045
Leu	His	Asn	Ala	Gln	Phe	Tyr	Leu	Leu	Gln	Arg	Thr	Pro	Glu	Val	Ala	
				275					280					285		
cg	agc	cg	gcc	acc	ccg	tta	tta	gat	ttg	atc	aag	aca	gcg	ttg	acg	1093
Arg	Ser	Arg	Ala	Thr	Pro	Leu	Leu	Asp	Leu	Ile	Lys	Thr	Ala	Leu	Thr	
			290					295					300			
ccc	cat	cca	ccg	caa	aaa	cag	gcg	tat	ggc	gtg	aca	tta	ccc	act	tca	1141
Pro	His	Pro	Pro	Gln	Lys	Gln	Ala	Tyr	Gly	Val	Thr	Leu	Pro	Thr	Ser	
		305					310					315				
gtg	ctg	ttt	atc	gcc	gga	cac	gat	act	aat	ctg	gca	aat	ctc	ggc	ggc	1189
Val	Leu	Phe	Ile	Ala	Gly	His	Asp	Thr	Asn	Leu	Ala	Asn	Leu	Gly	Gly	
	320					325					330					

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gca ctg gag ctc aac tgg acg ctt ccc ggt cag ccg gat aac acg ccg 1237
Ala Leu Glu Leu Asn Trp Thr Leu Pro Gly Gln Pro Asp Asn Thr Pro
335 340 345 350

cca ggt ggt gaa ctg gtg ttt gaa cgc tgg cgt cgg cta agc gat aac 1285
Pro Gly Gly Glu Leu Val Phe Glu Arg Trp Arg Arg Leu Ser Asp Asn
355 360 365

agc cag tgg att cag gtt tcg ctg gtc ttc cag act tta cag cag atg 1333
Ser Gln Trp Ile Gln Val Ser Leu Val Phe Gln Thr Leu Gln Gln Met
370 375 380

cgt gat aaa acg ccg ctg tca tta aat acg ccg ccc gga gag gtg aaa 1381
Arg Asp Lys Thr Pro Leu Ser Leu Asn Thr Pro Pro Gly Glu Val Lys
385 390 395

ctg acc ctg gca gga tgt gaa gag cga aat gcg cag ggc atg tgt tcg 1429
Leu Thr Leu Ala Gly Cys Glu Glu Arg Asn Ala Gln Gly Met Cys Ser
400 405 410

ttg gca ggt ttt acg caa atc gtg aat gaa gca cgc ata ccg gcg tgc 1477
Leu Ala Gly Phe Thr Gln Ile Val Asn Glu Ala Arg Ile Pro Ala Cys
415 420 425 430

agt ttg taatgcataa aaaagagcat tcagttacct gaatgctctg aggctgatga 1533
Ser Leu

caaacgaaga actgtctaat gcgtagaccg gaaaaggcgt tcacgccgca tccggccact 1593
ttcagttttc ctctttctcg gagtaactat aaccgtaata gttatagccg taactgtaag 1653
cggtgctggc gcgtttaatc acaccattga ggatagcgcc tttaatatg acgcctgcct 1713
gttccagacg ctgcattgac aaactcacct ctttgccggt gttcaagcca aaacgcgcaa 1773
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gcggcgtatc gacaatcacc agatcgtaaat ggctcgttcgc ccattccagt aattgacgca 1893
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<210> 8

<211> 432

<212> PRT

<213> Escherichia coli

<400> 8

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1 5 10 15
Pro Gln Ser Ala Phe Ala Gln Ser Glu Pro Glu Leu Lys Leu Glu Ser
20 25 30
Val Val Ile Val Ser Arg His Gly Val Arg Ala Pro Thr Lys Ala Thr
35 40 45
Gln Leu Met Gln Asp Val Thr Pro Asp Ala Trp Pro Thr Trp Pro Val
50 55 60
Lys Leu Gly Trp Leu Thr Pro Arg Gly Gly Glu Leu Ile Ala Tyr Leu
65 70 75 80
Gly His Tyr Gln Arg Gln Arg Leu Val Ala Asp Gly Leu Leu Ala Lys
85 90 95
Lys Gly Cys Pro Gln Ser Gly Gln Val Ala Ile Ile Ala Asp Val Asp
100 105 110
Glu Arg Thr Arg Lys Thr Gly Glu Ala Phe Ala Ala Gly Leu Ala Pro
115 120 125
Asp Cys Ala Ile Thr Val His Thr Gln Ala Asp Thr Ser Ser Pro Asp
130 135 140
Pro Leu Phe Asn Pro Leu Lys Thr Gly Val Cys Gln Leu Asp Asn Ala
145 150 155 160
Asn Val Thr Asp Ala Ile Leu Ser Arg Ala Gly Gly Ser Ile Ala Asp

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165
 Phe Thr Gly His Arg Gln Thr Ala Phe Arg Glu Leu Glu Arg Val Leu
 180 185 190
 Asn Phe Pro Gln Ser Asn Leu Cys Leu Lys Arg Glu Lys Gln Asp Glu
 195 200 205
 Ser Cys Ser Leu Thr Gln Ala Leu Pro Ser Glu Leu Lys Val Ser Ala
 210 215 220
 Asp Asn Val Ser Leu Thr Gly Ala Val Ser Leu Ala Ser Met Leu Thr
 225 230 235 240
 Glu Ile Phe Leu Leu Gln Gln Ala Gln Gly Met Pro Glu Pro Gly Trp
 245 250 255
 Gly Arg Ile Thr Asp Ser His Gln Trp Asn Thr Leu Leu Ser Leu His
 260 265 270
 Asn Ala Gln Phe Tyr Leu Leu Gln Arg Thr Pro Glu Val Ala Arg Ser
 275 280 285
 Arg Ala Thr Pro Leu Leu Asp Leu Ile Lys Thr Ala Leu Thr Pro His
 290 295 300
 Pro Pro Gln Lys Gln Ala Tyr Gly Val Thr Leu Pro Thr Ser Val Leu
 305 310 315 320
 Phe Ile Ala Gly His Asp Thr Asn Leu Ala Asn Leu Gly Gly Ala Leu
 325 330 335
 Glu Leu Asn Trp Thr Leu Pro Gly Gln Pro Asp Asn Thr Pro Pro Gly
 340 345 350
 Gly Glu Leu Val Phe Glu Arg Trp Arg Arg Leu Ser Asp Asn Ser Gln
 355 360 365
 Trp Ile Gln Val Ser Leu Val Phe Gln Thr Leu Gln Gln Met Arg Asp
 370 375 380
 Lys Thr Pro Leu Ser Leu Asn Thr Pro Pro Gly Glu Val Lys Leu Thr
 385 390 395 400
 Leu Ala Gly Cys Glu Glu Arg Asn Ala Gln Gly Met Cys Ser Leu Ala
 405 410 415
 Gly Phe Thr Gln Ile Val Asn Glu Ala Arg Ile Pro Ala Cys Ser Leu
 420 425 430

<210> 9

<211> 1308

<212> DNA

<213> Artificial Sequence

<220>

<223> modified phytase enzyme

<221> CDS

<222> (1)...(1296)

<400> 9

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1				5				10						15		

ccg	caa	tct	gca	ttc	gct	cag	agt	gag	ccg	gag	ctg	aag	ctg	gaa	agt	96
Pro	Gln	Ser	Ala	Phe	Ala	Gln	Ser	Glu	Pro	Glu	Leu	Lys	Leu	Glu	Ser	
			20					25					30			

ccg	caa	tct	gca	ttc	gct	cag	agt	gag	ccg	gag	ctg	aag	ctg	gaa	agt	96
Pro	Gln	Ser	Ala	Phe	Ala	Gln	Ser	Glu	Pro	Glu	Leu	Lys	Leu	Glu	Ser	
			20					25					30			

gtg	gtg	att	gtc	agt	cgt	cat	ggg	gtg	cgt	gct	cca	acc	aag	gcc	acg	144
Val	Val	Ile	Val	Ser	Arg	His	Gly	Val	Arg	Ala	Pro	Thr	Lys	Ala	Thr	
			35				40					45				

caa	ctg	atg	cag	gat	gtc	acc	cca	gac	gca	tgg	cca	acc	tgg	ccg	gta	192
Gln	Leu	Met	Gln	Asp	Val	Thr	Pro	Asp	Ala	Trp	Pro	Thr	Trp	Pro	Val	
			50			55					60					

aaa	ctg	ggg	gag	ctg	aca	ccg	cgc	ggg	ggg	gag	cta	atc	gcc	tat	ctc	240
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Lys 65	Leu	Gly	Glu	Leu	Thr 70	Pro	Arg	Gly	Gly	Glu 75	Leu	Ile	Ala	Tyr	Leu 80	
gga Gly	cat His	tac Tyr	tgg Trp	cgt Arg 85	cag Gln	cgt Arg	ctg Leu	gta Val	gcc Ala 90	gac Asp	gga Gly	ttg Leu	ctg Leu	cct Pro 95	aaa Lys	288
tgt Cys	ggc Gly	tgc Cys	ccg Pro 100	cag Gln	tct Ser	ggt Gly	cag Gln	gtc Val 105	gcg Ala	att Ile	att Ile	gct Ala 110	gat Asp	gtc Val	gac Asp	336
gag Glu	cgt Arg	acc Thr 115	cgt Arg	aaa Lys	aca Thr	ggc Gly	gaa Glu 120	gcc Ala	ttc Phe	gcc Ala	gcc Ala	ggg Gly 125	ctg Leu	gca Ala	cct Pro	384
gac Asp 130	tgt Cys	gca Ala	ata Ile	acc Thr	gta Val	cat His 135	acc Thr	cag Gln	gca Ala	gat Asp	acg Thr 140	tcc Ser	agt Ser	ccc Pro	gat Asp	432
ccg Pro 145	tta Leu	ttt Phe	aat Asn	cct Pro	cta Leu 150	aaa Lys	act Thr	ggc Gly	gtt Val	tgc Cys 155	caa Gln	ctg Leu	gat Asp	aac Asn	gcg Ala 160	480
aac Asn	gtg Val	act Thr	gac Asp	gcg Ala 165	atc Ile	ctc Leu	gag Glu	agg Arg	gca Ala 170	gga Gly	ggg Gly	tca Ser	att Ile	gct Ala 175	gac Asp	528
ttt Phe	acc Thr	ggg Gly	cat His 180	tat Tyr	caa Gln	acg Thr	gcg Ala	ttt Phe 185	cgc Arg	gaa Glu	ctg Leu	gaa Glu	cgg Arg 190	gtg Val	ctt Leu	576
aat Asn	ttt Phe	ccg Pro 195	caa Gln	tca Ser	aac Asn	ttg Leu	tgc Cys 200	ctt Leu	aaa Lys	cgt Arg	gag Glu	aaa Lys 205	cag Gln	gac Asp	gaa Glu	624
agc Ser 210	tgt Cys	tca Ser	tta Leu	acg Thr	cag Gln	gca Ala 215	tta Leu	cca Pro	tcg Ser	gaa Glu	ctc Leu 220	aag Lys	gtg Val	agc Ser	gcc Ala	672
gac Asp 225	tgt Cys	gtc Val	tca Ser	tta Leu	acc Thr 230	ggt Gly	gcg Ala	gta Val	agc Ser	ctc Leu 235	gca Ala	tca Ser	atg Met	ctg Leu	acg Thr 240	720
gag Glu	ata Ile	ttt Phe	ctc Leu	ctg Leu 245	caa Gln	caa Gln	gca Ala	cag Gln	gga Gly 250	atg Met	ccg Pro	gag Glu	ccg Pro	ggg Gly 255	tgg Trp	768
gga Gly	agg Arg	atc Ile	acc Thr 260	gat Asp	tca Ser	cac His	cag Gln	tgg Trp 265	aac Asn	acc Thr	ttg Leu	cta Leu	agt Ser 270	ttg Leu	cat His	816
aac Asn	gcg Ala	caa Gln 275	ttt Phe	gat Asp	ttg Leu	cta Leu	caa Gln 280	cgc Arg	acg Thr	cca Pro	gag Glu	gtt Val 285	gcc Ala	cgc Arg	agc Ser	864
cgc Arg 290	gcc Ala	acc Thr	ccg Pro	tta Leu	tta Leu	gat Asp 295	ttg Leu	atc Ile	aag Lys	aca Thr	gcg Ala 300	ttg Leu	acg Thr	ccc Pro	cat His	912
cca Pro 305	ccg Pro	caa Gln	aaa Lys	cag Gln	gcg Ala 310	tat Tyr	ggt Gly	gtg Val	aca Thr	tta Leu 315	ccc Pro	act Thr	tca Ser	gtg Val	ctg Leu 320	960

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ttt atc gcc gga cac gat act aat ctg gca aat ctc ggc ggc gca ctg 1008
 Phe Ile Ala Gly His Asp Thr Asn Leu Ala Asn Leu Gly Gly Ala Leu
 325 330

gag ctc aac tgg acg ctt ccc ggt cag ccg gat aac acg ccg cca ggt 1056
 Glu Leu Asn Trp Thr Leu Pro Gly Gln Pro Asp Asn Thr Pro Pro Gly
 340 345 350

ggt gaa ctg gtg ttt gaa cgc tgg cgt cgg cta agc gat aac agc cag 1104
 Gly Glu Leu Val Phe Glu Arg Trp Arg Arg Leu Ser Asp Asn Ser Gln
 355 360 365

tgg att cag gtt tcg ctg gtc ttc cag act tta cag cag atg cgt gat 1152
 Trp Ile Gln Val Ser Leu Val Phe Gln Thr Leu Gln Gln Met Arg Asp
 370 375 380

aaa acg ccg ctg tca tta aat acg ccg ccc gga gag gtg aaa ctg acc 1200
 Lys Thr Pro Leu Ser Leu Asn Thr Pro Pro Gly Glu Val Lys Leu Thr
 385 390 395 400

ctg gca gga tgt gaa gag cga aat gcg cag ggc atg tgt tcg ttg gca 1248
 Leu Ala Gly Cys Glu Glu Arg Asn Ala Gln Gly Met Cys Ser Leu Ala
 405 410 415

ggt ttt acg caa atc gtg aat gaa gca cgc ata ccg gcg tgc agt ttg 1296
 Gly Phe Thr Gln Ile Val Asn Glu Ala Arg Ile Pro Ala Cys Ser Leu
 420 425 430

agatctcatc ta 1308

<210> 10

<211> 432

<212> PRT

<213> Artificial Sequence

<220>

<223> modified phytase enzyme

<400> 10

Met Lys Ala Ile Leu Ile Pro Phe Leu Ser Leu Leu Ile Pro Leu Thr
 1 5 10 15
 Pro Gln Ser Ala Phe Ala Gln Ser Glu Pro Glu Leu Lys Leu Glu Ser
 20 25 30
 Val Val Ile Val Ser Arg His Gly Val Arg Ala Pro Thr Lys Ala Thr
 35 40 45
 Gln Leu Met Gln Asp Val Thr Pro Asp Ala Trp Pro Thr Trp Pro Val
 50 55 60
 Lys Leu Gly Glu Leu Thr Pro Arg Gly Gly Glu Leu Ile Ala Tyr Leu
 65 70 75 80
 Gly His Tyr Trp Arg Gln Arg Leu Val Ala Asp Gly Leu Leu Pro Lys
 85 90 95
 Cys Gly Cys Pro Gln Ser Gly Gln Val Ala Ile Ile Ala Asp Val Asp
 100 105 110
 Glu Arg Thr Arg Lys Thr Gly Glu Ala Phe Ala Ala Gly Leu Ala Pro
 115 120 125
 Asp Cys Ala Ile Thr Val His Thr Gln Ala Asp Thr Ser Ser Pro Asp
 130 135 140
 Pro Leu Phe Asn Pro Leu Lys Thr Gly Val Cys Gln Leu Asp Asn Ala
 145 150 155 160
 Asn Val Thr Asp Ala Ile Leu Glu Arg Ala Gly Gly Ser Ile Ala Asp
 165 170 175
 Phe Thr Gly His Tyr Gln Thr Ala Phe Arg Glu Leu Glu Arg Val Leu
 180 185 190

10306703.txt

Asn	Phe	Pro	Gln	Ser	Asn	Leu	Cys	Leu	Lys	Arg	Glu	Lys	Gln	Asp	Glu
		195					200					205			
Ser	Cys	Ser	Leu	Thr	Gln	Ala	Leu	Pro	Ser	Glu	Leu	Lys	Val	Ser	Ala
	210					215					220				
Asp	Cys	Val	Ser	Leu	Thr	Gly	Ala	Val	Ser	Leu	Ala	Ser	Met	Leu	Thr
225					230					235					240
Glu	Ile	Phe	Leu	Leu	Gln	Gln	Ala	Gln	Gly	Met	Pro	Glu	Pro	Gly	Trp
			245						250					255	
Gly	Arg	Ile	Thr	Asp	Ser	His	Gln	Trp	Asn	Thr	Leu	Leu	Ser	Leu	His
			260					265						270	
Asn	Ala	Gln	Phe	Asp	Leu	Leu	Gln	Arg	Thr	Pro	Glu	Val	Ala	Arg	Ser
		275					280					285			
Arg	Ala	Thr	Pro	Leu	Leu	Asp	Leu	Ile	Lys	Thr	Ala	Leu	Thr	Pro	His
	290					295					300				
Pro	Pro	Gln	Lys	Gln	Ala	Tyr	Gly	Val	Thr	Leu	Pro	Thr	Ser	Val	Leu
305					310					315					320
Phe	Ile	Ala	Gly	His	Asp	Thr	Asn	Leu	Ala	Asn	Leu	Gly	Gly	Ala	Leu
			325						330					335	
Glu	Leu	Asn	Trp	Thr	Leu	Pro	Gly	Gln	Pro	Asp	Asn	Thr	Pro	Pro	Gly
			340					345						350	
Gly	Glu	Leu	Val	Phe	Glu	Arg	Trp	Arg	Arg	Leu	Ser	Asp	Asn	Ser	Gln
		355					360					365			
Trp	Ile	Gln	Val	Ser	Leu	Val	Phe	Gln	Thr	Leu	Gln	Gln	Met	Arg	Asp
	370					375					380				
Lys	Thr	Pro	Leu	Ser	Leu	Asn	Thr	Pro	Pro	Gly	Glu	Val	Lys	Leu	Thr
385					390					395					400
Leu	Ala	Gly	Cys	Glu	Glu	Arg	Asn	Ala	Gln	Gly	Met	Cys	Ser	Leu	Ala
			405						410					415	
Gly	Phe	Thr	Gln	Ile	Val	Asn	Glu	Ala	Arg	Ile	Pro	Ala	Cys	Ser	Leu
			420					425					430		